

## Complete Genome Sequence of *Streptomyces cattleya* NRRL 8057, a Producer of Antibiotics and Fluorometabolites

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***Streptomyces cattleya*, a producer of the antibiotics thienamycin and cephamycin C, is one of the rare bacteria known to synthesize fluorinated metabolites. The genome consists of two linear replicons. The genes involved in fluorine metabolism and in the biosynthesis of the antibiotic thienamycin were mapped on both replicons.**

The bacterium *Streptomyces cattleya* has become an organism of interest due to its ability to produce various antibiotics (thienamycin, cephamycin C, penicillin N) (5) and to excrete the fluorinated antibiotic 4-fluorothreonine when cultivated in the presence of fluorine (11). *S. cattleya* has been used as a convenient model for characterizing both the enzyme responsible for the fluorination step and its substrate, as well as the metabolic pathway leading to 4-fluorothreonine (3, 4, 8).

The genome sequence was determined using a Sanger/pyrosequencing hybrid approach. Sanger reads (12-fold coverage, performed with libraries of 3-kb, 10-kb, and 25-kb fragments) were mixed with GS20 reads (13-fold coverage) (Roche Applied Science). For the assembly, the HybridAssemble version of Arachne (Broad Institute, www.broad.mit.edu) was used, and the assembly was validated with Consed (www.phrap.org) and the Mekano interface (Genoscope). The finishing step was achieved by primer walks and *in vitro* transposition. Coding sequence prediction and automatic annotation were performed using the Microscope platform (<http://www.genoscope.cns.fr/agc/microscope>) (12). Manual validations were carried out for specific genes.

The genome of *Streptomyces cattleya* NRRL 8057 is composed of one linear chromosome (SCAT, 6,283,062 bp, 72.94% GC content) and one linear megaplasmid (pSCAT, 1,809,491 bp, 73.21% GC content). Six rRNA operons, 64 tRNA genes, and 5,779 protein-coding genes were found on the chromosome, and 1,713 protein-coding genes were annotated on the plasmid.

The overall analysis of the gene content of the *S. cattleya* genome matches the features of a heterotrophic soil bacterium of the genus *Streptomyces*: a large supply of transporters, numerous catabolic enzymes for complex carbohydrates, multiple secondary metabolite biosynthesis genes, and versatile metabolic capabilities adapted to the complex environments and variable conditions where these organisms usually thrive (1).

The fluorometabolite biosynthesis pathway, as established

by the thorough studies of O'Hagan's group, is initiated by the fluorination of *S*-adenosylmethionine and leads to 4-fluorothreonine in four steps (3, 8). The previously cloned gene cluster (4) which contains the genes encoding the fluorinase and the 5'-fluoro-5'-deoxyadenosine phosphorylase was localized on the chromosome (SCAT\_4158 to SCAT\_4169). The isomerase gene involved in the formation of the intermediate 5-fluoro-5-deoxy-D-ribulose-1-phosphate (3, 9) was also located on the chromosome (SCAT\_2018). The gene for the aldolase that cleaves 5-fluoro-5-deoxy-D-ribulose-1-phosphate into 5-fluoroacetaldehyde and dihydroxyacetone-phosphate has not been identified yet in *S. cattleya*. SCAT\_1042 on the chromosome and SCAT\_p1154 and SCAT\_p1616 on the plasmid, annotated as fucose aldolases, are potential candidates. SCAT\_p0562 encodes the transaldolase responsible for the biosynthesis of 4-fluorothreonine from threonine and fluoroacetaldehyde (3). The published 17-amino-acid N-terminal sequence of the fluoroacetaldehyde dehydrogenase (6) perfectly matches SCAT\_0945, which was annotated as a probable aldehyde dehydrogenase gene.

The gene cluster involved in thienamycin biosynthesis (7, 10) is located on the plasmid (SCAT\_p0820 to SCAT\_p0840). The putative cephamycin gene cluster identified on the chromosome (SCAT\_5676 to SCAT\_p5692) has an organization similar to the one described for *Nocardia lactamdurans* (2). Candidate genes for resistance to virginiamycin and tylosin (SCAT\_p0715 and SCAT\_p0716, respectively) were identified. The chloramphenicol acetyltransferase gene is a pseudogene in *S. cattleya* (SCAT\_p1266-1267).

**Nucleotide sequence accession numbers.** The chromosome and megaplasmid sequences of *S. cattleya* NRRL8057 were deposited in EMBL under accession numbers FQ859185 and FQ859184, respectively. The sequences and the annotations are also available from the Microscope platform (<http://www.genoscope.cns.fr/agc/microscope>).

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